

WHAT IS CLAIMED IS:

1. A chimeric gene comprising a first nucleotide sequence comprising a 5' regulatory region operably linked to a nucleic acid sequence which encodes a regulator polypeptide and an untranslated 3' termination sequence, and a second nucleotide sequence comprising a 5' regulatory region operably linked to a nucleic acid sequence which is a coding or non-coding sequence, the expression of said nucleic acid sequence of said second nucleotide sequence being controlled by said regulator polypeptide of said first nucleotide sequence using an inducer, said inducer thereby causing modulation of expression of said nucleic acid sequence of said second nucleotide sequence, and said nucleotide sequence of said regulator polypeptide and/or said 5' regulatory region or parts thereof of said second nucleotide sequence being isolated from a prokaryote source.
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2. A chimeric gene according to Claim 1, wherein one or more of said 5' regulatory 10 regions each comprises a promoter which allows expression in eukaryote cells and/or tissues.
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3. A chimeric gene according to Claim 1 or 2, wherein the promoter of said 5' regulatory region operably linked to said nucleic acid encoding said regulator polypeptide is a constitutive, developmentally regulated, tissue-specific, cell-specific or cell compartment-specific promoter.
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4. A chimeric gene according to Claim 3, wherein said constitutive promoter is the CaMV 35S or CaMV 19S promoter.
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5. A chimeric gene according to Claim 3, wherein said tissue-specific promoter is the patatin promoter or the *petE* promoter.
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6. A chimeric gene according to Claim 3, wherein said cell compartment promoter is a chloroplast gene promoter or a mitochondrial gene promoter.
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7. A chimeric gene according to Claim 6, wherein said chloroplast gene promoter is of the gene encoding the large subunit of ribulose biphosphate carboxylase.
8. A chimeric gene according to Claim 6, wherein said mitochondrial gene promoter is of the 18S-5S rRNA gene.

9. A chimeric gene according to any one of Claims 1-8, wherein any one of said 5' regulatory regions comprises one or more enhancer sequences.

10. A chimeric gene according to Claim 9, wherein said enhancer sequence is a 5' transcriptional and/or translational enhancer sequence or an intron.

11. A chimeric gene according to Claim 10, wherein said enhancer is a non-translated leader sequence.

10 12. A chimeric gene according to Claim 11, wherein said non-translated leader sequence is a viral non-translated leader sequence.

13. A chimeric gene sequence according to Claim 12, wherein said enhancer is a non-translated leader sequence from the group consisting of Tobacco Mosaic Virus (TMV), 15 Maize Chlorotic Mottle Virus (MCMV), Alfalfa Mosaic Virus (AMV), Picornavirus, Potyvirus, AMV RNA4.

14. A chimeric gene sequence according to Claim 11, wherein said enhancer is the HSP 70 leader.

20 15. A chimeric gene according to Claim 10, wherein said enhancer is a transcriptional enhancer.

16. A chimeric gene according to Claim 15, wherein said enhancer is the *petE* enhancer.

25 17. A chimeric gene according to Claim 10, wherein said enhancer is an intron of the maize *Adh1* gene or the *Hsp70* intron from maize.

18. A chimeric gene according to Claim 1, wherein said regulator polypeptide comprises 30 one or more domains, which domain is a ligand binding domain, a nucleic acid binding domain, a transactivation domain, a silencing/repressing domain, a dimerisation domain or a targeting domain.

19. A chimeric gene according to Claim 18, wherein said ligand binding domain 35 suitably comprises a sequence of amino acids whose structure binds non-covalently to a complementary ligand.

20. A chimeric gene according to Claim 19, wherein said complementary ligand is said inducer, or a derivative or a precursor of said inducer.

21. A chimeric gene according to Claim 18, wherein said nucleic acid binding domain 5 comprises a sequence of amino acids which binds non-covalently to a response element.

22. A chimeric gene according to Claim 21, wherein said response element is located in the 5' regulatory region of said second nucleotide sequence.

10 23. A chimeric gene according to Claim 21, wherein said response element is a combination of two or more response elements responsive to one or more nucleic acid binding proteins.

24. A chimeric gene according to Claim 22, wherein said response element is selected 15 from the group consisting of response elements responding to LexA, Gal4, LacI, Tet, C1 or Ace1 proteins.

25. A chimeric gene according to Claim 18, wherein said transactivation domain is from Vp16 (isolated from the herpes simplex virus), from the maize C1 or a silencing/repressing 20 domain from the rice Oshox1 or the KRAB domain.

26. A chimeric gene according to Claim 18, wherein said targeting domain is one of the group consisting of plasma membrane, golgi, endoplasmatic reticulum, nuclear targeting signals, chloroplast, mitochondrial or inner envelope targeting sequences. 25

27. A chimeric gene according to any one of Claims 18-26, wherein said nucleotide sequence which encode any of the above domains are modified for improved expression in eukaryotes.

30 28. A chimeric gene according to Claim 18, wherein said regulator polypeptide comprises multiple domains of the same type.

29. A chimeric gene according to Claim 18, wherein said regulator polypeptide comprises a ligand binding domain and/or a DNA binding domain. 35

30. A chimeric gene according to Claim 18, wherein said regulator polypeptide is encoded by the nucleotide sequence from 295-1035bp of SEQ ID NO:1, sub-sequences thereof having the necessary function, or nucleic acid sequences substantially similar thereto that hybridize under low stringency conditions.

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31. A chimeric gene according to Claim 18, wherein said 5' regulatory region of the second nucleotide sequence comprises a core or full-length promoter sequence and the response element necessary for complementary binding of said regulator polypeptide of said first nucleotide sequence.

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32. A chimeric gene according to Claim 31, wherein said core promoter is the A1 core promoter obtained from the A1 gene of maize, the CaMV35S core promoter the CaMV35S full-length promoter or the CERV promoter.

15 33. A chimeric gene according to Claim 31, wherein said response element of said 5' regulatory region of said second nucleotide sequence is derived from the nucleotide sequence at 295-2805bp of SEQ ID NO: 1.

34. A chimeric gene according to Claim 33, wherein said response element is a 20 sequence isolated from the ohpA promoter region (1036-1260bp of SEQ ID NO: 1), sub-sequences of this sequence having the necessary function or substantially similar sequences thereto that hybridize under low stringency conditions.

35. A chimeric gene according to Claim 31, 32 or 34, wherein said sequence, sub-25 sequence or multiples thereof are arranged in normal or reverse orientation, upstream or downstream of the core or full-length promoter, and in any order thereof.

36. A chimeric gene according to Claim 30, wherein said coding sequence in the second nucleotide sequence is one or more of the sequences which encode proteins involved in 30 carbon metabolism; flowering; fertility and/or sterility; cell wall metabolism; genes that respond to environmental signals, such as pathogen attack; bacterium, fungus, virus, or insect resistance; or genes that confer resistance to antibiotics, herbicides or other toxic compounds.

35 37. A chimeric gene according to Claim 36, wherein said coding sequence is barnase or diphtheria toxin A-chain.

38. A method of controlling eukaryotic gene expression comprising introducing into a eukaryotic cell with an inducible gene expression system, said inducible gene expression system comprising a first nucleotide sequence comprising a 5' regulatory region operably linked to a nucleic acid sequence which encodes a regulator polypeptide and an untranslated 5' termination sequence, and a second nucleotide sequence comprising a 5' regulatory region operably linked to a nucleic acid sequence which is a coding or non-coding sequence, the expression of said nucleic acid sequence of said second nucleotide sequence being controlled by the regulator polypeptide of the first nucleotide sequence using an inducer, said inducer thereby causing modulation of expression of said nucleic acid 5

10 sequence of said second nucleotide sequence, and said nucleotide sequence of said regulator polypeptide and/or said 5' regulatory region, or parts thereof, of said second nucleotide sequence being isolated from a prokaryote source.

39. A method according to Claim 38, wherein said inducible gene expression system is 15 a chemically inducible gene expression system.

40. A method according to Claim 38 or 39, wherein said coding sequence is homologous or heterologous in origin with respect to the eukaryote being transformed.

20 41. A method according to Claim 38 or 39, wherein expression of said nucleic acid sequence of said second nucleotide sequence, said second nucleotide sequence being a target gene, is increased or decreased, whether from a basal or medial level respectively, or completely repressed or activated.

25 42. A method according to Claims 38 or 39, wherein an increase in target gene expression levels is caused by the addition or presence of said inducer.

43. A method according to Claims 38 or 39, wherein an increase in target gene expression levels is caused by the withdrawal or absence of said inducer.

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44. A method according to Claims 38 or 39, wherein a decrease in target gene expression levels is caused by the addition or presence of the inducer.

45. A method according to Claims 38 or 39, wherein a decrease in target gene expression levels is caused by the withdrawal or absence of the inducer.

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46. A method according to Claim 38 or 39, wherein, when said regulator polypeptide is encoded by the nucleotide 295-1053 sequence of SEQ ID NO: 1, said inducer is a chemical compound.

5 47. A method according to Claim 46, wherein said chemical compound is OHP, 2-hydroxy cinnamic acid, benzene, toluene, n-hexadecane or a functional equivalent of either.

48. A method according to Claim 38, wherein said inducer is a protein or nucleic acid sequence appropriate to a complementary domain of said regulator polypeptide.

10 49. A method according to Claim 38, wherein said 5' regulatory region of said second nucleotide sequence comprises one or more response elements, each being necessary for complementary binding of an appropriate domain or other portion of said regulator polypeptide.

15 50. A method according to Claim 38, wherein said inducer acts by indirect action.

51. A method according to Claim 38, wherein said inducer acts by direct action.

20 52. A method according to Claim 38, wherein said eukaryotic cell is a plant cell.

53. A method according to Claim 52, wherein said plant cell is from a monocot or dicot crop, a tree or other plant.

25 54. A method according to Claim 53, wherein said plant cell is a cell from one or more from the group consisting of potato, wheat, maize, barley, tomato, rice, canola, sugarbeet, tobacco, eucalyptus, populus, malus; or *Arabidopsis*.

55. A method according to Claims 38, wherein said gene expression system comprises a

30 single construct containing said first nucleotide sequence and said second nucleotide sequence.

56. A method according to Claims 38, wherein said gene expression system utilises two or more separate constructs.

57. A method according to Claim 56, wherein each construct is introduced into separate eukaryotes, which constructs are then transferred into one eukaryote by biological mating or crossing to bring the constructs together.

5 58. A method according to Claim 56, wherein said expression system comprises one transformation step followed by a further transformation step or steps.

59. Plant tissue transformed in accordance with the method of Claims 38.

10 60. The plasmid deposited under NCIMB Accession No. 40997.

61. A method of using the ohpA region of nucleotide 1036-1260 of SEQ ID NO: 1 or part thereof as the complementary response element to a DNA binding domain in eukaryotic cells.

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